



# Effective Ballast Water Treatment Solutions

# Hyde Marine, Inc.

## Ballast Water Treatment

- SeaKleen®
- Aquionics UV
- Arkal Filters
- Hyde Mud Remover
- New Technical Solutions



# SeaKleen®

## A Natural Biocide

- Developed by Garnett, Inc. - Atlanta
- Manufactured by Vitamar, Inc. – Memphis
- Extensive Testing by Univ. of Maryland, Chesapeake Biological Laboratory – Drs. Rodger Dawson and David Wright.
- Full-scale Efficacy Testing Aboard Cape May in Summer of 2001 including Bioassays to Confirm No Residual Toxicity.
- Testing Approved by MD Dept. of Natural Resources.





# Garnett, Inc

Watkinsville, Georgia USA

## SeaKleen®

The Natural Solution to Ballast Water Treatment™



# SeaKleen®

## A Natural Biocide

- Toxicity testing on bi-valves completed and toxicity testing on herring planned for Washington State
- Planned dilution study in Puget Sound
- Additional Testing by Univ. of Maryland, Chesapeake Biological Laboratory in 2003
- Full-scale Efficacy Testing Aboard an ITB tanker in Puget Sound expected in Summer of 2003 by Univ. of Wash. scientists..



# SeaKleen®

## A Natural Biocide

- Toxic to Broad Range of Organisms in Both Fresh and Salt Water
- Lethal to Dinoflagellate Cysts
- 98% Effective at 1 ppm – 100% at 2 ppm
- Low Toxicity to Mammals, Birds, and Higher Species of Fish – Vitamin K3
- Short Half-life – Degrades to Harmless Products Within Days



# Hyde's Full Scale Shipboard System Experience

## UV Treatment with Cyclonic Separation:

- April 2000 – “Regal Princess” – 200 m<sup>3</sup>/hr
- Aug. 2001 – “Sea Princess” – 220 m<sup>3</sup>/hr
- Mar. 2002 – “R. J. Pfeiffer” – 350 m<sup>3</sup>/hr
- Feb. 2002 – “Star Princess” – 255 m<sup>3</sup>/hr
- Apr. 2002 – “Stolt Aspiration” – 250m<sup>3</sup>/hr

## UV Treatment with Filtration Pretreatment:

May 2003 – “Coral Princess” – 250m<sup>3</sup>/hr



# Hyde's Testing Experience

- Great Lakes Ballast Technology Demonstration Project Barge Testing
  - 1998 Screen Filtration
  - 2000 Screen Filtration and Cyclonic Separation with UV
  - 2001 Arkal Disk Filtration and UV
- 2000 Full Scale Testing on “Regal Princess”
- 2002 & 2003 Full Scale Testing on Newer Installations
- 2001 “Cape May” Full Scale Testing of UV with Filtration and of Seakleen.
- 2003 & 2004 Full Scale Test Programs Expected on UV with Filtration and SeaKleen.





# Treatment System Requirements

## Applies to Any Treatment Technology:

- Maximize killing, inactivation or removal of living organisms from the ballast water.
- Meet demands of the shipboard marine environment.
- Minimize adverse effects on environment.
- Meet the existing safety standards of the marine industry, regulatory bodies and the vessel operating company.



# SeaKleen® Lab Test Results

- 2ppm Destroyed Dinoflagellate cysts within 2h
- 1ppm bleached alga *Isochysis galbana* <24h
- 0.5ppm bleached alga *Neochloris* sp. <24h
- Toxic to freshwater amphipods  $\leq 1.5\text{ppm}$
- 100% toxicity to West Coast mussels (*Mytilus galloprovincialis*) at 1ppm after 24h
- *E-coli* growth inhibition (Kirby-Bauer test) at 1ppm



# SeaKleen® Lab Test Results

- Toxic to zebra mussel larvae at 500ppb and above after 24h
- Sheephead Minnow eggs & larvae killed at 1ppm (24h)
- 1ppm toxic to *Vibrio fischeri* (congeneric with cholera)
- Toxic to “red tide” components including *Gymnodinium brevi* at 500ppb
- Toxic to freshwater ostracod crustacean *Daphnia magna* at 1.5ppm



# SeaKleen®

## Shipboard Test “Cape May”



# SeaKleen®

## Shipboard Test Results

(Cape May, Baltimore Harbor summer 2001)

- Complete Mortality of all zooplankton after 24 hours at 2ppm (not tested at lower concentrations)
- Phytoplankton growth arrested and chloroplasts bleached after 24 hours



# Treatment of Cape May Ballast Tanks and Mesocosms with Seakleen<sup>®</sup> 100:0<sup>1</sup> at 2 ppm

## Mesocosm Population After 24 hours Exposure

Zooplankton	% Alive	% Mortality
Rotifers	0	100
Polychaetae	N/F	N/F
Copepod	0	100
Copepodites	0	100
Nauplii	0	100
Bivalve larvae	0	100

<sup>1</sup> 100:0 represents 100% menadione sodium bisulfite  
N/F = Not Found



# Treatment of Cape May Ballast Tanks and Mesocosms with Seakleen® 100:0<sup>1</sup> at 1 ppm

Zooplankton	Mesocosm Population After 24 hours Exposure		Mesocosm Population After 48 hours Exposure	
	% Alive	% Mortality	% Alive	%Mortality
Rotifers	90	10	2	98
Polychaetae	100	0	6	94
Copepod	50	50	0	100
Copepodites	69	31	0	100
Nauplii	94	6	0	100
Bivalve larvae	95	5	6	94

<sup>1</sup> 100:0 represents 100% menadione sodium bisulfite



# Mussel Larvae Test of Seakleen<sup>®</sup>

Mussel (*Mytilus galloprovincialis*) larvae test using Static 48-Hour Exposure to Seakleen<sup>®</sup> 80:20 and 0:100 under differing light exposure regimes <sup>1,2</sup>

Test Solutions aged for 48 hours in either Total Darkness or under Light Conditions

<sup>1</sup> x:y are x = Menadione Sodium Bisulfite and y = Menadione Wettable Powder

<sup>2</sup> Based on average initial count of 247 embryos per 10 ml sample





# Mussel Larvae (*Mytilus galloprovincialis*)

## Test of Seakleen® 80:20

Test Solutions Aged 48 hours in Total Darkness  
Bioassay Conducted under Total Darkness

Concentration ppm	% Mortality
0.5	100**
0.2	0
0.1	0
0.05	0
Control	0

\*\*  $p \leq 0.05$



# Mussel Larvae (*Mytilus galloprovincialis*)

## Test of Seakleen® 80:20

Test Solutions Aged 48 hours in Total Darkness  
Bioassay Conducted under Constant Light

Concentration ppm	% Mortality	
0.5	100**	
0.2	7.3	
0.1	1.6	** $p \leq 0.05$
0.05	0	
Control	0	



# Mussel Larvae (*Mytilus galloprovincialis*)

## Test of Seakleen® 80:20

Test Solutions Aged 48 hours in Constant Light  
Bioassay Conducted under Constant Light

Concentration ppm	% Mortality	
0.5	100**	
0.2	4.3	
0.1	0	** $p \leq 0.05$
0.05	0	
Control	0	



# Mussel Larvae (*Mytilus galloprovincialis*)

## Test of Seakleen® 0:100

Test Solutions Aged 48 hours in Total Darkness  
Bioassay Conducted under Total Darkness

Concentration ppm	% Mortality	
0.2	100**	
0.15	25.3**	
0.05	1.7	** $p \leq 0.05$
0.02	0	
Control	0	



# Mussel Larvae (*Mytilus galloprovincialis*)

## Test of Seakleen® 0:100

Test Solutions Aged 48 hours in Total Darkness  
Bioassay Conducted under Constant Light

Concentration ppm	% Mortality	
0.2	94.4**	
0.15	3.7**	
0.05	0	** $p \leq 0.05$
0.02	0	
Control	0	



# Mussel Larvae (*Mytilus galloprovincialis*)

## Test of Seakleen® 0:100

Test Solutions Aged 48 hours in Constant Light  
Bioassay Conducted under Constant Light

Concentration ppm	% Mortality	
0.2	69.9**	
0.15	8.0**	
0.05	0	** $p \leq 0.05$
0.02	0	
Control	0	

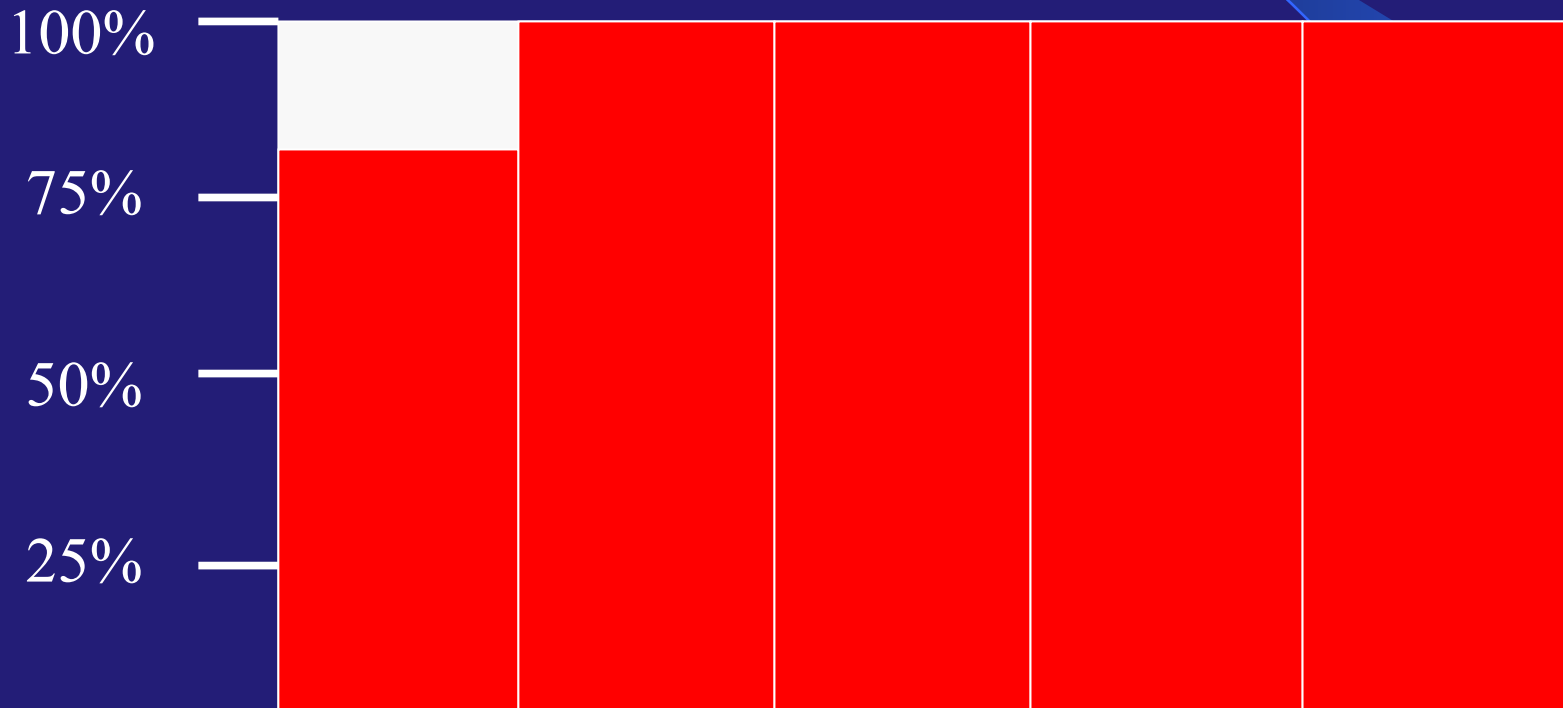


# Dinoflagellates

## Effects of Menadione against *Glenodium*

Cell  
Death

2 ppm



Time

2 Hours

24 Hours

48 Hours

72 Hours

96 Hours

80%

100%

100%

100%

100%

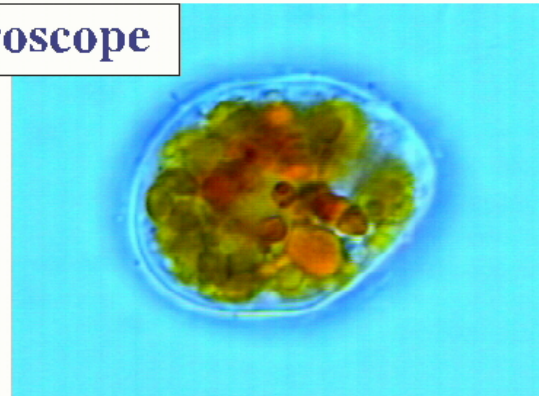
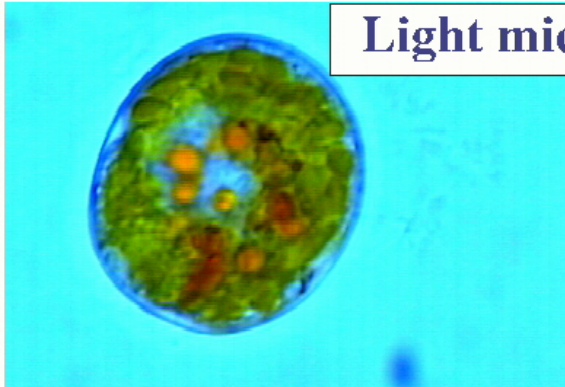
# *Glenodinium foliaceum* cysts

## 2 Hours after Exposure to Seakleen®

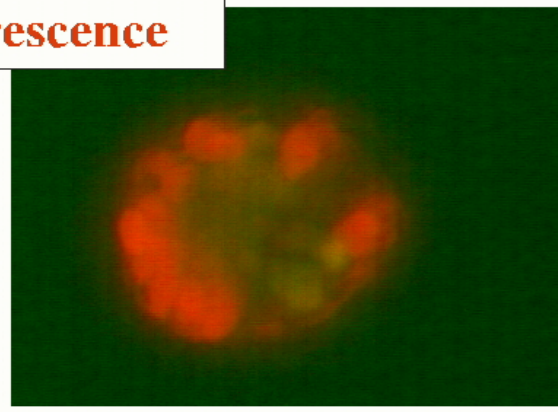
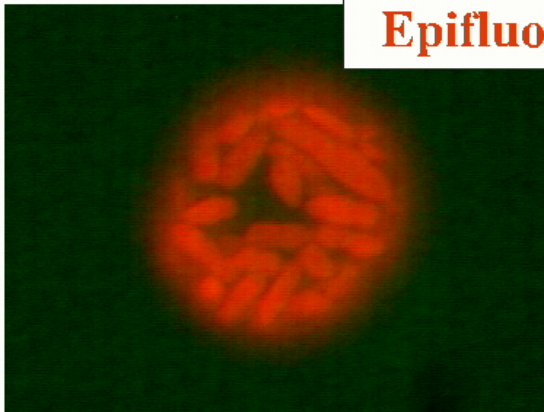
**Control  
(No SK®)**

**Treated with  
Seakleen® 2 ppm**

Light microscope

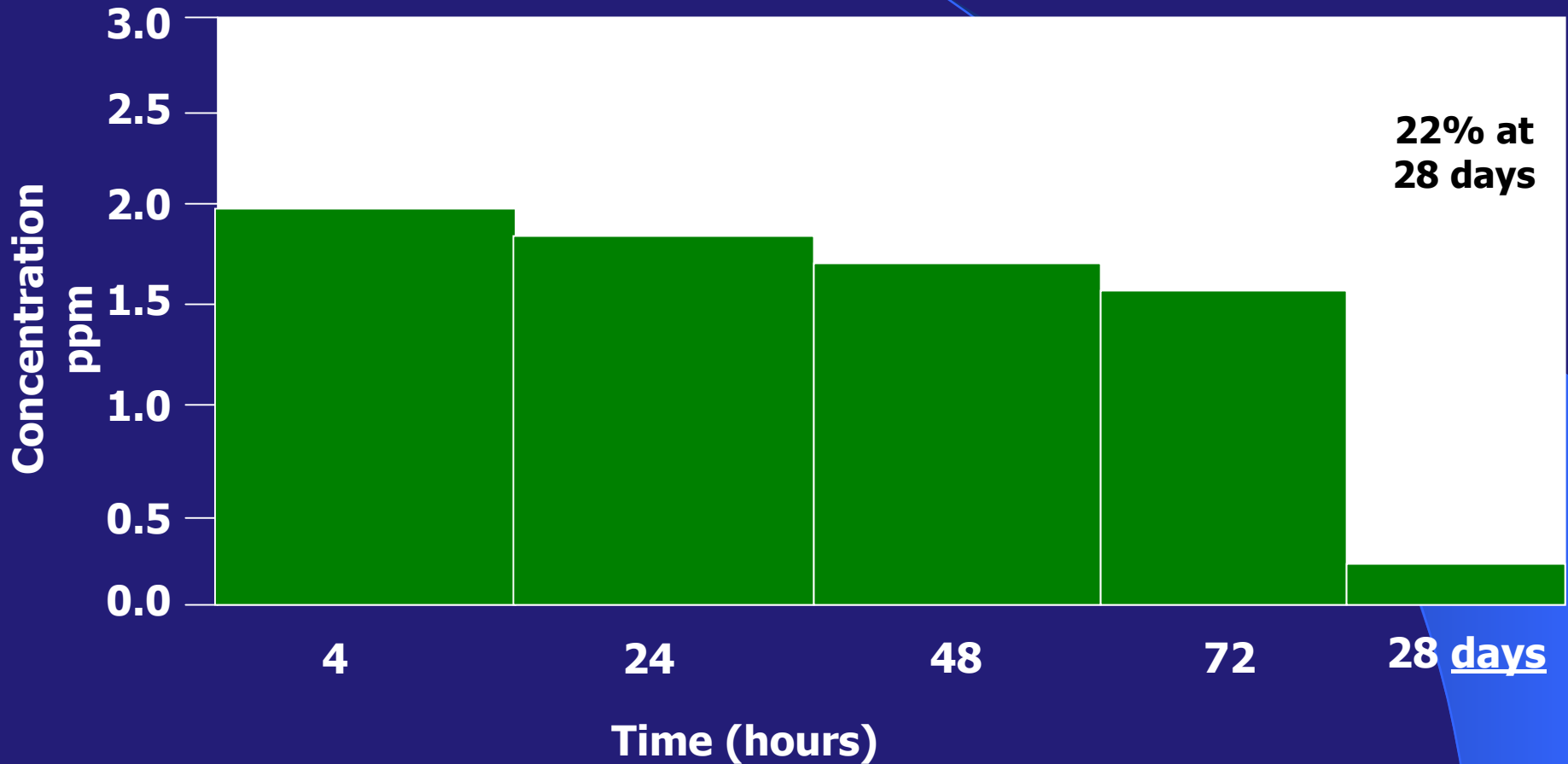


Epifluorescence

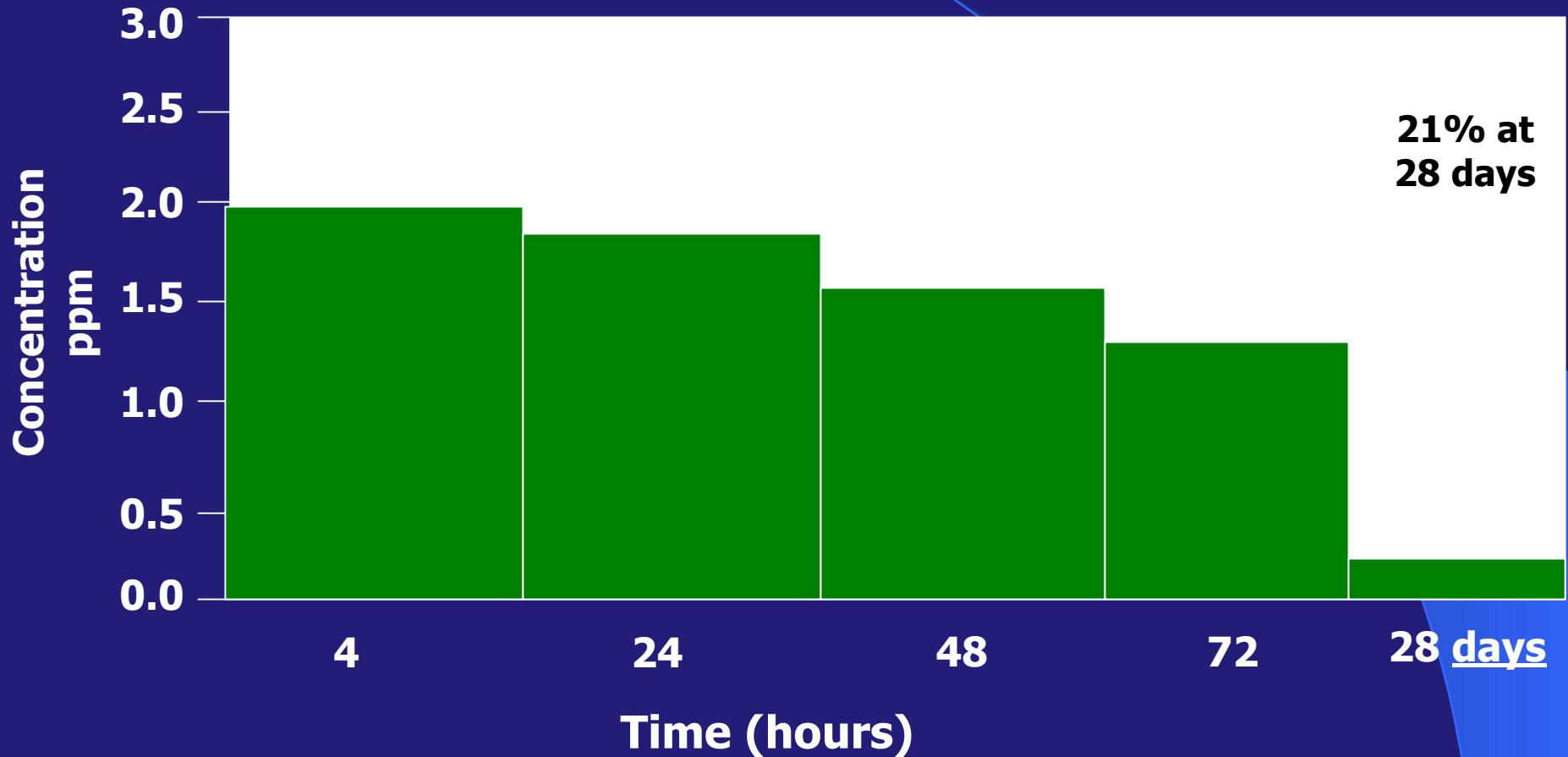




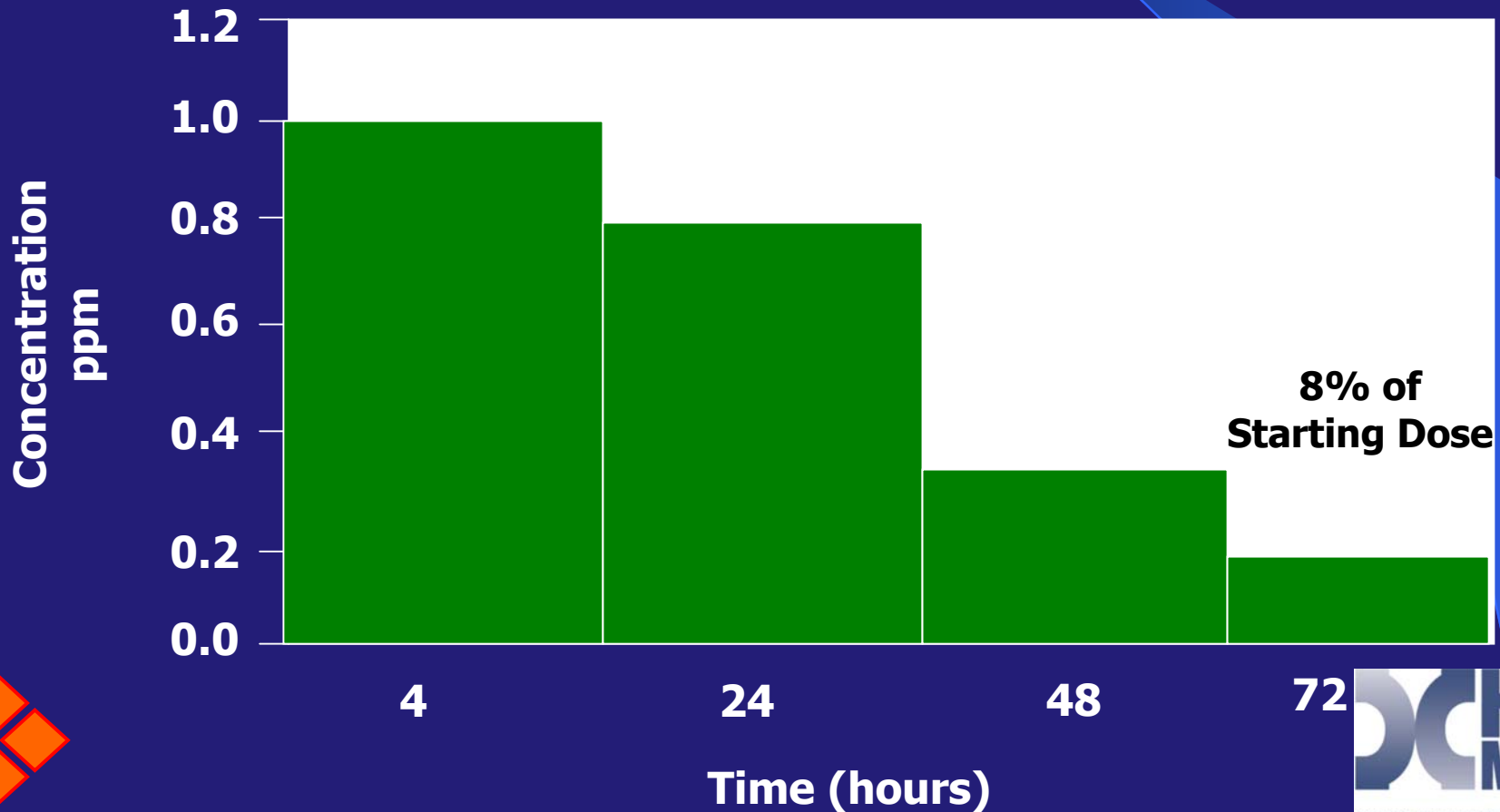
# HPLC (High Performance Liquid Chromatography) Analysis of SeaKleen® in River Water Exposed to Darkness



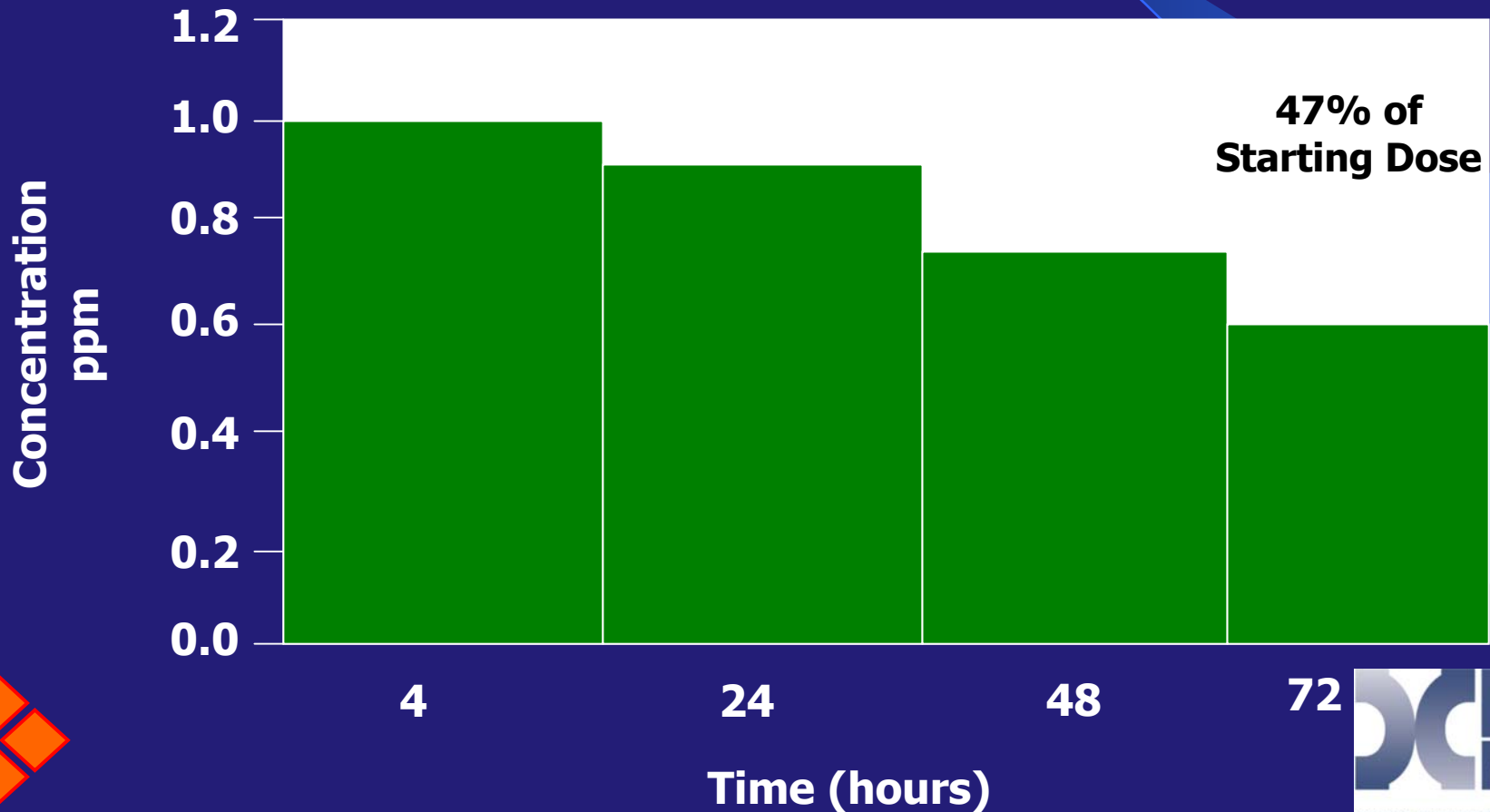
# HPLC Analysis of SeaKleen® in Sea Water Exposed to Darkness



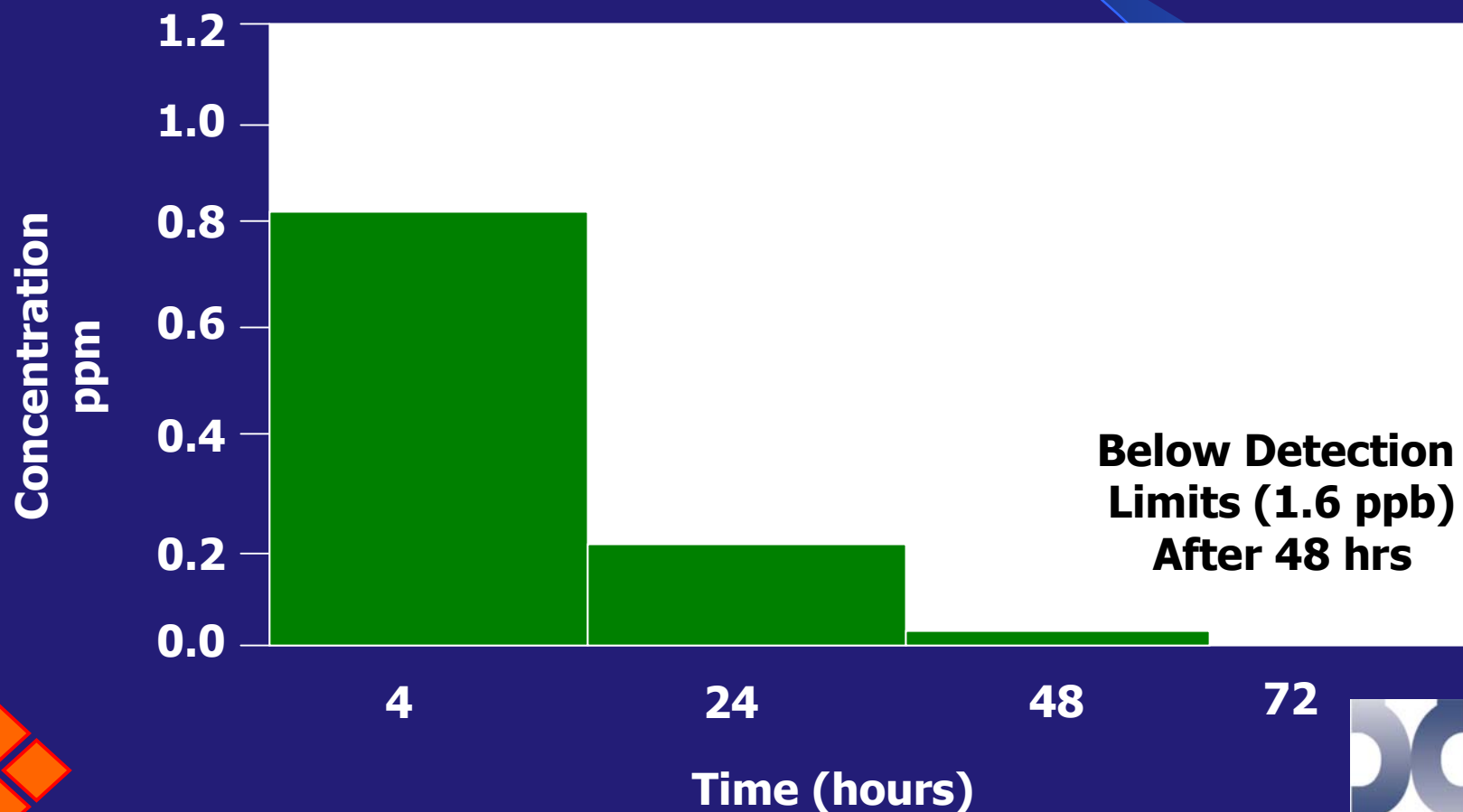
# HPLC Analysis of SeaKleen® in River Water Exposed to Sunlight



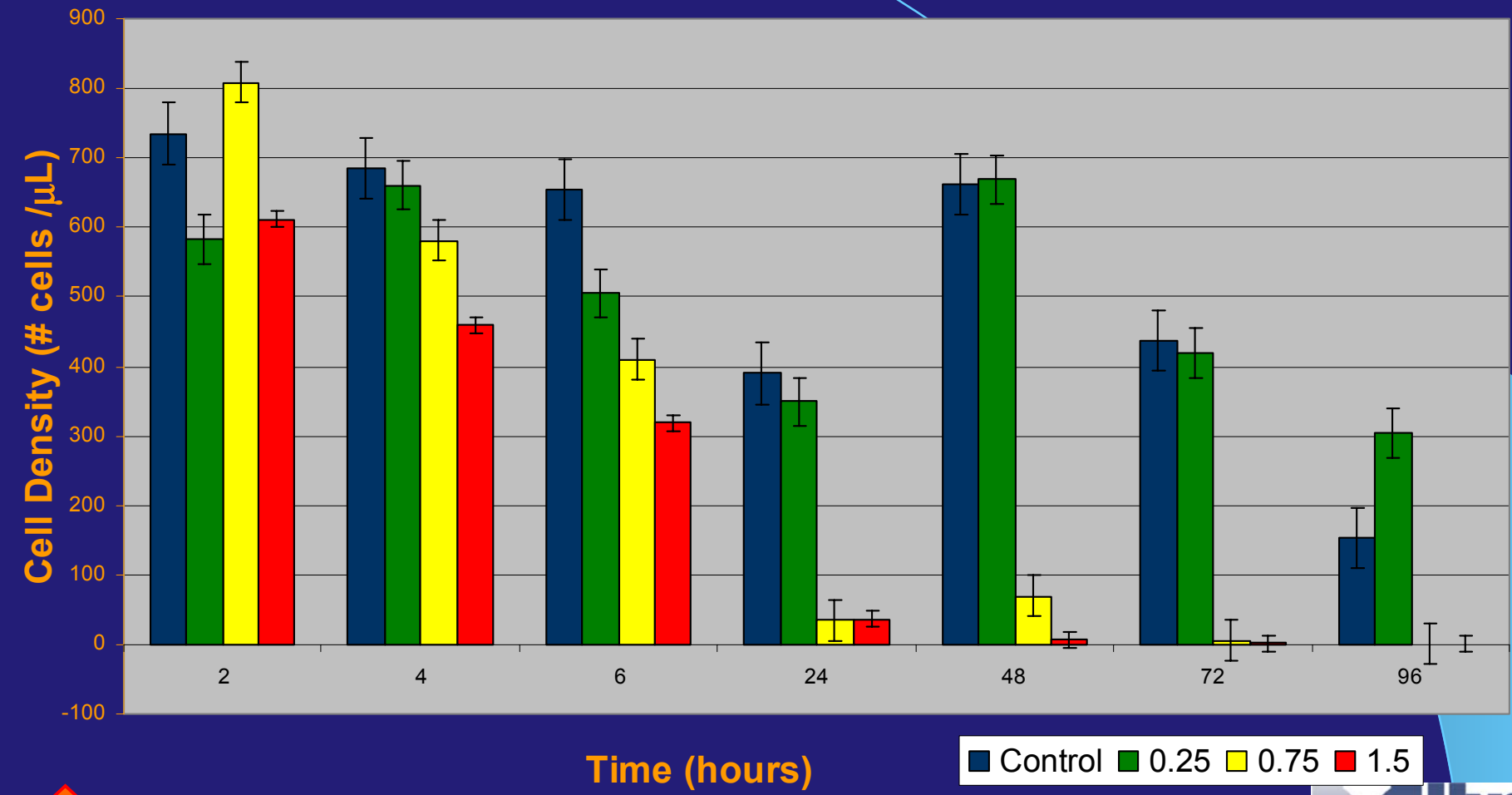
# HPLC Analysis of SeaKleen® in Sea Water Exposed to Sunlight



# HPLC Analysis of SeaKleen® in River Water Exposed to Aquatic Organisms (i.e., Blue-Green Algae)



## *Isochrysis galbana* SeaKleen Study



# SeaKleen®

## A Natural Biocide

- **Cost Effective** – 15 to 20 cents per tonne
- **Highly Soluble** in Fresh and Salt Water
- **Not Corrosive** to Piping or Ballast Tanks - Unlike Oxidizing Biocides
- Delivered in **Safe Solid** Form – Can be Stored on Board and Handled by Crew With No Special Training
- Low Affinity for Particulate Matter and Sediment



# SeaKleen®

## A Natural Biocide

- Particularly Suitable for Bulk Carriers and Tankers with Large Ballast Volumes
- Testing Planned Aboard Operating Tankers and RRF Ships in Baltimore Harbor
- EPA Registration Pending
- Straightforward Dosing Under all Ballasting Conditions Including Gravity Ballasting
- Intelligent Dosing Systems with Minimum Installation Costs





# Hyde Marine - What's Next?

- Continuing Testing and Development of **SeaKleen®** ongoing in 2003 & 2004
- Adapting New Technologies, such as Med. Press UV & Disk Filtration, to Improve Performance of **SeaKleen®**
- Completion of Registration Process and Commercial Introduction of **SeaKleen®**
- Hyde Marine is Committed to Continuous Improvement of Ballast Water Treatment Technologies

